

AMENDMENT TO THE SPECIFICATION

Please delete the previously submitted sequence listing and the sequence listing from the English translation of the application and replace it with the sequence listing submitted on compact disc enclosed herewith.

In the specification at page 1, after the paragraph of Related Applications added in the Preliminary Amendment, please insert the following:

-- INCORPORATION OF SEQUENCE LISTING

The contents of the following submission on compact discs are incorporated herein by reference in its entirety: two copies of the Sequence Listing (COPY 1 and COPY 2) and a computer-readable form of the Sequence Listing (CRF COPY), all on CD-ROMs, each containing: file name: Replacement Sequence list-13195-00006-US, date recorded: May 29, 2007, size: 128 KB.

FIELD OF THE INVENTION --

In the specification at page 1 before line 5, before the paragraph starting with "Amino acids...," please insert the following heading:

-- BACKGROUND --

In the specification at page 5 after line 12, before the paragraph starting with "It is an object" please insert the following heading:

-- BRIEF SUMMARY OF THE INVENTION --

In the specification at page 5 after line 24, before the paragraph starting with "Threonine-degrading proteins...," please insert the following new paragraphs and headings:

- Advantageously, there is introduction in process step (a) a nucleic acid sequence selected from the group
- i) of a nucleic acid sequence having the sequence depicted in SEQ ID NO: 1; SEQ ID NO: 11; SEQ ID NO: 13; SEQ ID NO: 15; SEQ ID NO: 17; SEQ ID NO: 19; SEQ ID NO: 21; SEQ ID NO: 23 or SEQ ID NO: 25;
 - ii) of a nucleic acid sequence obtained owing to the degeneracy of the genetic code through back-translation of the amino acid sequence depicted in SEQ ID NO: 2, SEQ ID NO: 12; SEQ ID NO: 14; SEQ ID NO: 16; SEQ ID NO: 18; SEQ ID NO: 20; SEQ ID NO: 22; SEQ ID NO: 24 or SEQ ID NO: 26 and
 - iii) of a derivative of the nucleic acid sequence depicted in SEQ ID NO: 1, SEQ ID NO: 11; SEQ ID NO: 13; SEQ ID NO: 15; SEQ ID NO: 17; SEQ ID NO: 19; SEQ ID NO: 21; SEQ ID NO: 23 or SEQ ID NO: 25; which codes for a polypeptide having at least 50% homology at the amino acid level with the amino acid sequence depicted in SEQ ID NO: 2, SEQ ID NO: 12; SEQ ID NO: 14; SEQ ID NO: 16; SEQ ID NO: 18; SEQ ID NO: 20; SEQ ID NO: 22; SEQ ID NO: 24 or SEQ ID NO: 26 with a negligible reduction in the biological activity of the polypeptides.

BRIEF DESCRIPTION OF THE FIGURES

Figure 1 represents the consensus of threonine aldolase which are able to be used in the process of the invention. The following amino acid sequences are presented: YEL046c (SEQ ID NO: 33); P1;T24108 (SEQ ID NO: 34); Q87I10 (SEQ ID NO: 35); GLY1_YEAST (SEQ ID NO: 36); P1;T38302 (SEQ ID NO: 37); P1;E75410 (SEQ ID NO: 38); Q9VCK6 (SEQ ID NO: 39); Q885J1 (SEQ ID NO: 40); P1;G83533 (SEQ ID NO: 41); Q83S08 (SEQ ID NO: 42); P1;F64825 (SEQ ID NO: 43); P1;AF0608 (SEQ ID NO: 44); Q87HF4 (SEQ ID NO: 45); P1;E82418 (SEQ ID NO: 46); P1;T46877 (SEQ ID NO: 47); Q9M835 (SEQ ID NO: 48); Q8RCY7 (SEQ ID NO: 49); P1;C72215 (SEQ ID NO: 50); Q896G8 (SEQ ID NO: 51);

P1;C84060 (SEQ ID NO: 52); Q89N26 (SEQ ID NO: 53); Q9X8S4 (SEQ ID NO: 54); P1;D84395 (SEQ ID NO: 55); CAA02484 (SEQ ID NO: 56); TOXG_COCCA (SEQ ID NO: 57); and P1;AF1474 (SEQ ID NO: 58). The consensus sequence is shown in SEQ ID NO: 59.

Figure 2 represents the consensus of the lysine-degrading protein which are able to be used in the process of the invention. The following amino acid sequences are listed in Figure 2 as numbers (1., 2., 3., etc.) which are shown in square brackets as follows: YJL055w [1] (SEQ ID NO: 60); Q871Q6 [2] (SEQ ID NO: 61); Q815T3 [3] (SEQ ID NO: 62); Q81XE4 [4] (SEQ ID NO: 63); P1;D70033 [5] (SEQ ID NO: 64); Q8H7U8 [6] (SEQ ID NO: 65); Q8L8B8 [7] (SEQ ID NO: 66); Q8XXM6 [8] (SEQ ID NO: 67); Q88DF4 [9] (SEQ ID NO: 68); P1;A83031 [10] (SEQ ID NO: 69); Q8PAJ9 [11] (SEQ ID NO: 70); Q8PMA0 [12] (SEQ ID NO: 71); Q8NN34 [13] (SEQ ID NO: 72); P1;AI3438 [14] (SEQ ID NO: 73); Q8G289 [15] (SEQ ID NO: 74); Q984W8 [16] (SEQ ID NO: 75); P1;B97490 [17] (SEQ ID NO: 76); P1;B83993 [18] (SEQ ID NO: 77); Q8A2T1 [19] (SEQ ID NO: 78); Q92R13 [20] (SEQ ID NO: 79); Q8ETC2 [21] (SEQ ID NO: 80); Q8NXQ6 [22] (SEQ ID NO: 81); Q8CTK0 [23] (SEQ ID NO: 82); P1;F55578 [24] (SEQ ID NO: 83); Q839D0 [25] (SEQ ID NO: 84); P1;D84035 [26] (SEQ ID NO: 85); Q8EZ03 [27] (SEQ ID NO: 86); Q89NP4 [28] (SEQ ID NO: 87); and Q8RFZ1 [29] (SEQ ID NO: 88). The consensus sequence is shown in SEQ ID NO: 29.

DETAILED DESCRIPTION OF THE INVENTION --

In the specification at page 6, line 6, please replace the paragraph starting with “An advantageous embodiment” with the following amended paragraph:

An advantageous embodiment of the process of the invention is a process for preparing amino acids, advantageously L-methionine, in transgenic organisms, which process comprises the following steps:

- a) introduction of a nucleic acid sequence which codes for a threonine-degrading protein which comprises the following consensus sequence

H[x]₂G[X]R[X]₁₉D[X]₇K[X]₂₇G (SEQ ID NO: 27), or

HDGAR[X]₃A[X]₁₅D[X]₄CXSK[X]₄PXGS[X]₃G[X]₇A[X]₄K[X]₂GGGXRQXG (SEQ ID NO: 28)

- b) introduction of a nucleic acid sequence which increases the threonine degradation in the transgenic organism, and
- c) expression of a nucleic acid sequence mentioned under (a) or (b) in the transgenic organism.

In the specification at page 7, line 24, please replace the paragraph starting with “One advantageous embodiment” with the following amended paragraph:

One advantageous embodiment of the process of the invention is a process for preparing amino acids, advantageously L-methionine, in transgenic organisms, which process comprises the following steps:

- a) introduction of a nucleic acid sequence which codes for a lysine-degrading protein which comprises the following consensus sequence
*G[X]4GIM[X]45M[X]2RK[X]2M[X]1,GGXG[X]3E[X]2E[X]3W (SEQ ID NO: 29), or
LG[X]9LVYGG[X]3GIMGXVA[X]9G[X]3GXIP[X]24MHXRK[X]2M[X]6F[X]3PGGXGTXEE[X]2
E[X]2TW[X]2IG[X]3KP[X]4N[X]3FY[X]14F (SEQ ID NO: 30)*
- b) introduction of nucleic acid sequence which increases the lysine degradation in the transgenic organism, and
- c) expression of a nucleic acid sequence mentioned under (a) or (b) in the transgenic organism.

In the specification at page 8, line 31, please replace the paragraph starting with “In a further embodiment” with the following amended paragraph:

In a further embodiment of the process, it is a process for preparing amino acids, advantageously L-methionine, in transgenic organisms, which process comprises the following steps:

- a) introduction of a nucleic acid sequence which codes for a threonine-degrading protein which comprises the following consensus sequence
*H[x]2G[X]R[X]19D[X]7K[X]27G (SEQ ID NO: 27), or
HXdGAR[X]3A[X]15D[X]4CXSK[X]4PXGS[X]3G[X]7A[X]4K[X]2GGGXRQXG (SEQ ID NO: 28)*
- and introduction of a nucleic acid sequence which codes for a lysine-degrading protein which comprises the following consensus sequence

G[X]4GIM[X]45M[X]2RK[X]2M[X]11GGXG[X]3E[X]2E[X]3W (SEQ ID NO: 29), or

LG[X]9LVYGG[X]3GIMGXVA[X]9G[X]3GXIP[X]24MHXRK[X]2M[X]6F[X]3PGGXGTXEE[X]2
E[X]2TW[X]2IG[X]3KP[X]4N[X]3FY[X]14F (SEQ ID NO: 30), or

- b) introduction of a nucleic acid sequence which codes for proteins which increase threonine degradation and lysine degradation in the transgenic organisms, and
- c) expression of a nucleic acid sequence mentioned under (a) or (b) in the transgenic organism.

In the specification at page 41, line 14, please replace the paragraph starting with "The nucleic acid molecules" with the following amended paragraph:

The nucleic acid molecules used in the process, e.g. a nucleic acid molecule having a nucleotide sequence of SEQ ID NO: 1, SEQ ID NO: 11, SEQ ID NO: 13, SEQ ID NO: 15, SEQ ID NO: 17, SEQ ID NO: 19, SEQ ID NO: 21, SEQ ID NO: 23, or SEQ ID NO: 25 or of a part thereof, can be isolated by use of standard techniques of molecular biology and the sequence information provided herein. It is also possible with the aid of comparison algorithms to identify for example a homologous sequence or homologous, conserved sequence regions at the DNA or amino acid level. These can be used as hybridization probe as well as standard hybridization techniques (as described, for example, in Sambrook et al., Molecular Cloning: A Laboratory Manual, 2nd edition, Cold Spring Harbor Laboratory, Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY, 1989) for isolating further nucleic acid sequences useful in the process. Moreover, a nucleic acid molecule comprising a complete sequence of SEQ ID NO: 1, SEQ ID NO: 11, SEQ ID NO: 13, SEQ ID NO: 15, SEQ ID NO: 17, SEQ ID NO: 19, SEQ ID NO: 21, SEQ ID NO: 23, or SEQ ID NO: 25 or a part thereof can be isolated by polymerase chain reaction using oligonucleotide primers based on this sequence or parts thereof (e.g. a nucleic acid molecule comprising the complete sequence or a part thereof can be isolated by polymerase chain reaction using oligonucleotide primers constructed on the basis of this same sequence). For example, mRNA can be isolated from cells (e.g. by the guanidinium thiocyanate extraction process of Chirgwin et al. (1979) Biochemistry 18:5294-5299) and cDNA can be prepared using reverse transcriptase (e.g. Moloney MLV reverse transcriptase obtainable from Gibco/BRL, Bethesda, MD, or AMV reverse transcriptase obtainable from Seikagaku America, Inc., St. Petersburg, FL). Synthetic oligonucleotide primers for amplification using the polymerase chain reaction can be designed on the basis of one of the amino acid sequences depicted in SEQ ID NO: 1, SEQ ID NO: 11, SEQ ID NO: 13, SEQ ID NO: 15, SEQ ID NO: 17, SEQ ID NO: 19, SEQ ID NO: 21, SEQ ID NO: 23, or SEQ ID NO: 25 or with the aid of the amino acid sequences depicted in SEQ ID NO: 2, SEQ ID NO: 12, SEQ ID NO: 14, SEQ ID NO: 16, SEQ ID NO: 18,

SEQ ID NO: 20, SEQ ID NO: 22, SEQ ID NO: 24, or SEQ ID NO: 26. A further possibility is to identify, by protein sequence comparisons of threonine aldolases or lysine decarboxylases from various organisms, conserved regions from which in turn degenerate primers can then be derived. Such degenerate primers may be derived from the consensus sequences

H[x]₂G[X]R[X]₁₉D[X]₇K[X]₂₇G (SEQ ID NO: 27),
HXdGAR[X]₃A[X]₁₅D[X]₄CXSK[X]₄PXGS[X]₃G[X]₇A[X]₄K[X]₂GGGXRXQXG (SEQ ID NO: 28),
G[X]₄GIM[X]₄₅M[X]₂RK[X]₂M[X]₁₁GGXG[X]₃E[X]₂E[X]₃W (SEQ ID NO: 29), or
LG[X]₉LVYGG[X]₃GIMGXVA[X]₉G[X]₃GXIP[X]₂₄MHXRK[X]₂M[X]₆F[X]₃PGGXGTXEE[X]₂
E[X]₂TW[X]₂IG[X]₃KP[X]₄N[X]₃FY[X]₁₄F (SEQ ID NO: 30). These degenerate primers can then be utilized for amplifying fragments of new threonine aldolases and/or lysine decarboxylases from other organisms by PCR. These fragments can then be utilized as hybridization probe for isolating the complete gene sequence. An alternative possibility is to isolate the missing 5' and 3' sequences by means of RACE-PCR. A nucleic acid of the invention can be amplified using cDNA or, alternatively, genomic DNA as template and suitable oligonucleotide primers in standard PCR amplification techniques. The nucleic acid amplified in this way can be cloned into a suitable vector and characterized by DNA sequence analysis. Oligonucleotides corresponding to a nucleotide sequence used in the process can be prepared by standard synthetic processes, for example using an automatic DNA synthesizer.

In the specification at page 60, line 10, please replace the paragraph starting with "The primer sequences" with the following amended paragraph:

The following primer sequences were chosen for the gene of SEQ ID NO: 1:

- i) forward primer (SEQ ID NO: 1) (SEQ ID NO: 31)

5'-GGAATTCCAGCTGACCACCATGACTGAATTCGAATTGCCTCCAA

- ii) reverse primer (SEQ ID NO: 1) (SEQ ID NO: 32)

5'-GATCCCCGGGAATTGCCATGTCAGTATTGTAGGTTTTATTCGC